REMARKS

Claims 1, 3-10, 12-19, 21-23 and 25-32 are in this application.

Claims 18 has been amended to insert – a – before "potting".

Therefore, the claim objection is moot.

Claims 2, 11, and 24 have been cancelled. Claim 1 has been amended to include the subject matter of claim 24.

According to the Examiner, claims 1, 6-8, 10, 12-17, 19, 21-24, 28, and 30-32 remain rejected under 35 USC 103(a) as being unpatentable over Mishra et al (Plant Cell, Tissue and Organ Culture 73:21-35, 2003) in view of Dasgupta et al (US 2005/0235377 A1). These rejections are again respectfully traversed.

No combination of these references teach or make obvious the claimed process. The instant invention relates to synchronized regeneration of cotton plants using tissue culture techniques (see ¶0008, ¶0010, ¶0019, ¶0022, ¶0091, ¶0109, and Table 10). As shown in Table 10 and as explained in the paragraph on page 25 following Table 10:

> when embryogenic mass were subcultured to inositol free basal medium for a single cycle of 10 days and then returned to basal medium containing inositol, it produced a synchronized development of somatic embryos (100% embryos in globular stage after one subculture to basal medium; 91.66% embryos in heart stage after 2 subcultures to basal medium and 81.99% embryos in torpedo stage after 3 subcultures). The total number of mature embryos produced per explant was increased to a 4-5 fold higher value. The embryogenic clumps, when subcultured to an inositol-free medium for 2 cycles showed a reduction in homogeneity in the development stages of embryos. Hence, single cycle of inositol starvation was required for inducing a high level of synchrony in embryogenesis.

This is consistent with claim 1 and supports that use of an inositol-free medium for 8-12 days before the globular stage results in synchronized growth.

There is no suggestion or motivation in the references or in the art to combine Mishra et al with Dasgupta because the asynchronous embryonic development described in Mishra teaches away from the substantially synchronized development of the present invention.

Mishra et al. discloses a media composition containing myo-inositol for callus induction media and regeneration of cotton from hypocotyl explant. Mishra et al. also carries out experiments to study the effect of various hormone combinations of auxins and cytokinins. Mishra et al. do not teach withholding of myo-inositol in embryogenesis media for starvation, and then releasing myo-inositol for synchronized growth of embryos. This is the novel and inventive feature of the present invention. Mishra et al. reports that development of embryos is asynchronous but provides no solution as how they can be made synchronous (see col. 2 on page 25). This prior art problem has been solved by the present invention.

Since Mishra teaches a feature and result directly contrary to those of claim 1 and the dependent claims thereof, there would be no motivation for a person having ordinary skill in the art to utilize that reference in conceiving the present invention.

The disclosure in Dasgupta et al. relates to a genetic engineering method for the development of stress tolerant plants. Different media compositions (MSAg, CC-, CC-2, Delay, Selection, Regeneration and Plant development) have been used and for different purpose like MSAg media for initial phase, co-cultivation media for transformation, selection media for selection of transformed cells, regeneration media for regeneration of transformed embryos (Table 3). The nutrient composition is different for these media based on the objective for which it was used (para 0121, 0123, 0125; and Table 3). In the regeneration media of Dasgupta, et al. myo-inositol is used for regenerating plants from the embryos and not for synchronized growth after starvation of embryogenic tissues as in present invention. Starvation at a particular stage is done to bring all the tissues at same stage growth, i.e. globular stage, and then the stress factor is released by supplying it in the media so that all of them reach to maturity at the same time, i.e. synchronized growth. In Dasgupta, the purpose of withholding inositol and the supplying it again after selection of transformed embryos for regeneration is

not disclosed.

Starvation of embryogenic tissues for inositol before globular stage, for 8-12 days and thereafter adding it for synchronized growth and further development, is not taught by Dasgupta et al.

The explant used by Dasgupta is immature embryos or immature seeds (para 0121) and not hypocotyl, mesocotyl or cotyledon pieces of the present invention and as included in claim 1 of this aplication. The embryos were cultured in MSAg media, then transferred to co-cultivation media and then cells was transferred to selection media (without myo-inositol) where the calli was left for 2-3 months to attain the size of 10mm and then transferred to regeneration (with myo-inositol) media (para 0123 and para 0125). This is contrasted with the present invention, where the explants cultured are hypocotyls, mesocotyl or cotyledon pieces, these de-differentiates into callus, which is then starved by withdrawing inositol for synchronized growth before the formation of matured somatic enbryos. Thereafter, mature somatic embryos were transferred to regeneration media with inositol in it.

In Dasgupta et al. inositol was present only in regeneration media and not in other media compositions used before regeneration (Table 3). In the present invention, inositol was present in all the media composition (germination, callus induction, embryogenesis and regeneration) except for 8-12 days in embryogenesis induction medium.

The Examiner's contention set on pages 2 and 3 of the action that the end result i.e. development of plantlets is the same in the instant invention as that of Dasgupta et al. is erroneous. The novel and inventive feature of the present invention lies in the synchronized growth of the embryos and high frequency (4-5 fold) of mature embryos per explant were obtained from globular embryos (table 10, pages 25-26) after the inositol starvation step.

The Examiner's remark on pages 3 and 4 of the action that Dasgupta et al. teaches that calli developed into plants deprived of inositol therefore synchronized embyro-developments is the effect of intended use is incorrect. In Dasgupta et al. calli the media used for development into plants had inositol. The citation does not disclose withholding of inositol for

synchronized embryogenesis. The Examiner's statement on page 4 of the Office Action that Dasgupta et al. showed that no inositol is necessary until the calli reached 10 mm size is incorrect, as the document is silent on this aspect.

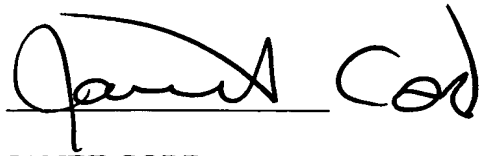
The Examiner's statement that the instant claims do not cite increased embryogenesis when explants are deprived on inositol is not correct as support for increased embryogenesis is present as described above, on pages 24-26 of the specification and is Example 6, and Table 1.

Dasgupta et al. (para 0125-0128) do not teach synchronization by inositol deprivation, instead they selected their transgenic calli on selection medium, where selection agent is hygromycin. Hygromycin helps in selecting transgenic callus and not in attaining synchronization. Mishra et al. also did not obtain synchronized embryos. In fact, they reported that "development of somatic embryos at all stages could be easily observed at this time (page 25, stage VII, Fig 2). Therefore, the combination of Dasgupta et al, and Mishra et al. do not make obvious the claimed invention, which is regeneration of cotton plants by short-term inositol deprivation to attain synchronous embryos. Synchronous embryos help in reducing the time for regeneration because all the embryos are obtained at the same time as and the number of embryos recovered is high.

Accordingly, it is respectfully requested that this rejection be withdrawn.

Applicants respectfully submit that the application is now in allowable form. An early notice of allowance is earnestly solicited and is believed to be fully warranted.

Respectfully submitted,



JANET CORD
LADAS & PARRY LLP
26 WEST 61ST STREET
NEW YORK, NEW YORK 10023
REG. NO. 33778
(212)708-1935